MOLECULAR CELL BIOLOGY
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W.H. FREEMAN AND COMPANY

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Molecular Cell Biology ➡ **3. Protein Structure and Function** ➡ 3.5. Purifying, Detecting, and Characterizing Proteins

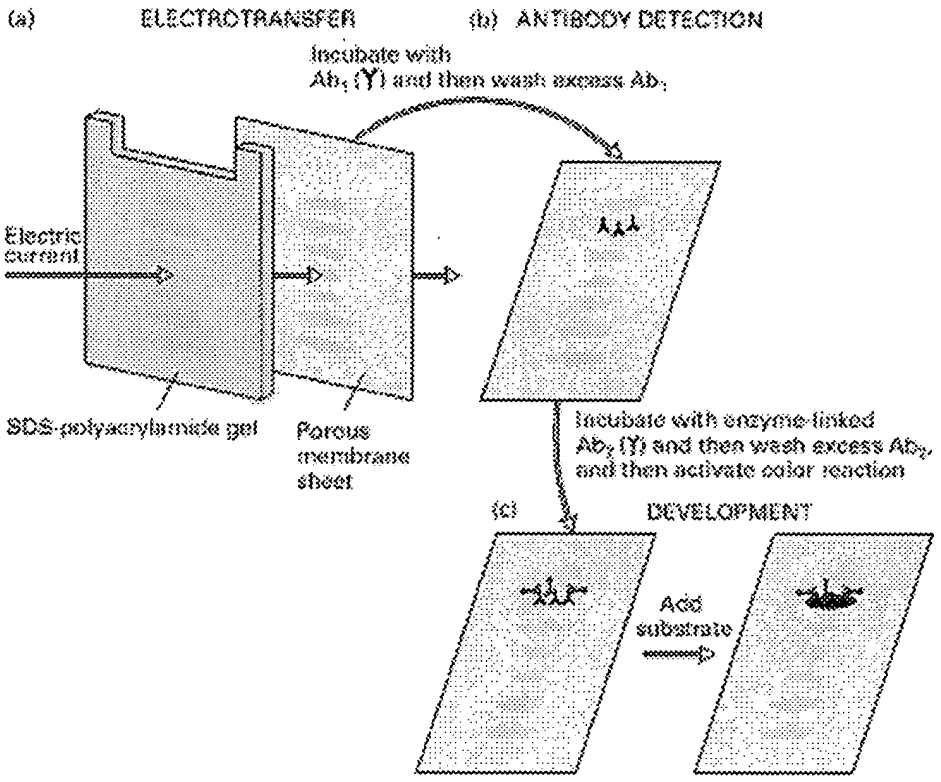

Figure 3-44. Western blotting, or immunoblotting. (a) A protein mixture is electrophoresed through an SDS gel, and then transferred from the gel onto a membrane. (b) The membrane is flooded with a solution of antibody (Ab_1) specific for the desired protein. Only the band containing this protein binds the antibody, forming a layer of antibody molecules (although their position can't be seen at this point). After sufficient time for binding, the membrane is washed to remove unbound Ab_1. (c) In the development step, the membrane first is incubated with a second antibody (Ab_2) that binds to the bound Ab_1. This second antibody is covalently linked to alkaline phosphatase, which catalyzes a chromogenic reaction. Finally, the substrate is added and a deep purple precipitate forms, marking the band containing the desired protein.

Immunoblotting.

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